Apr 4th, 4:45 PM - 5:00 PM

# The effects of diluted bitumen (dilbit) exposure during embryonic development on the future swimming performance and metabolic and ionic recovery post-exercise in sockeye salmon (Oncorhynchus nerka)

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# The effects of diluted bitumen on the embryonic development and swimming performance of sockeye salmon (*Oncorhynchus nerka*)

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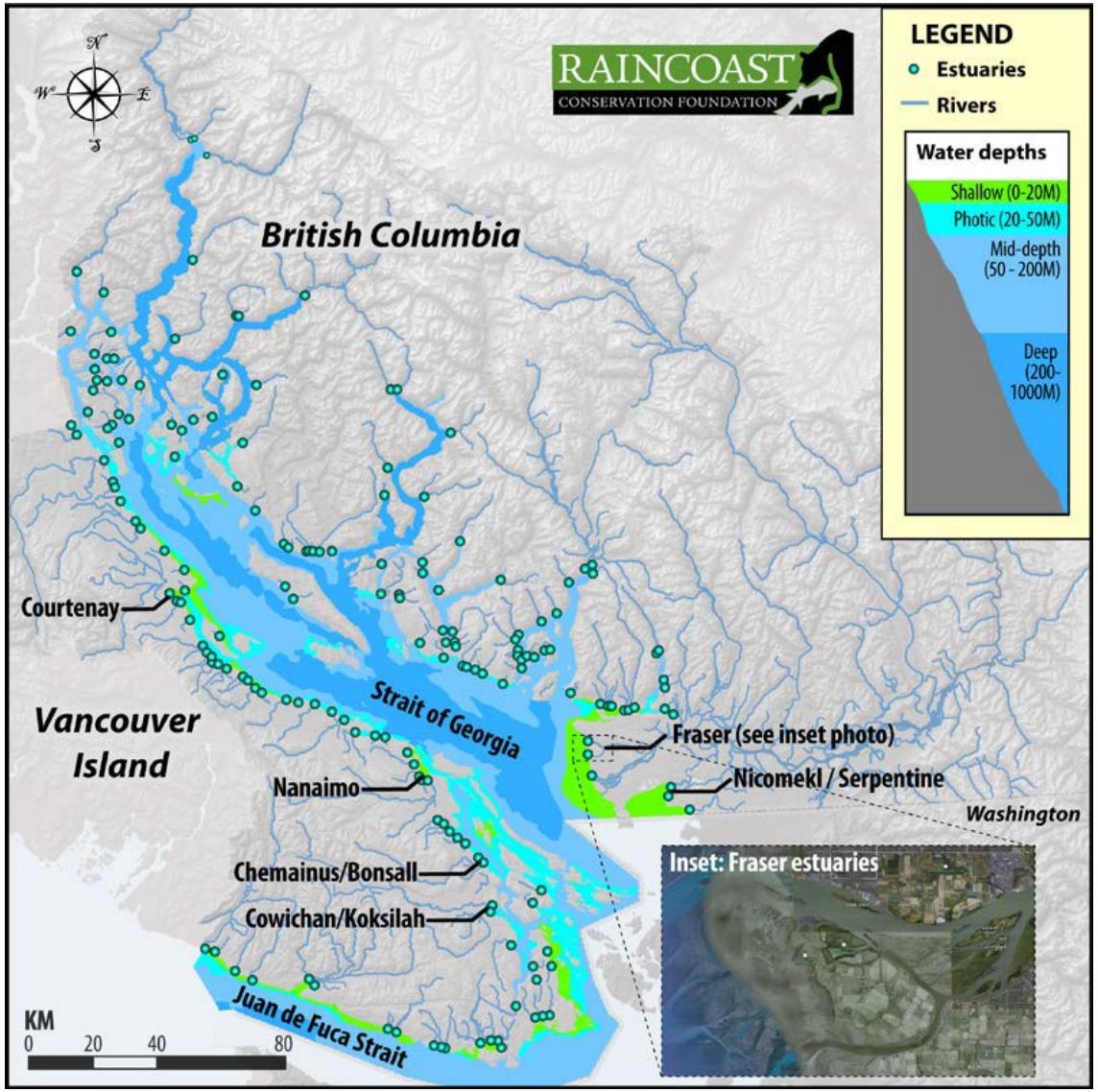
30th Salish Sea Ecosystem Conference, April 4-6, 2018

# Background



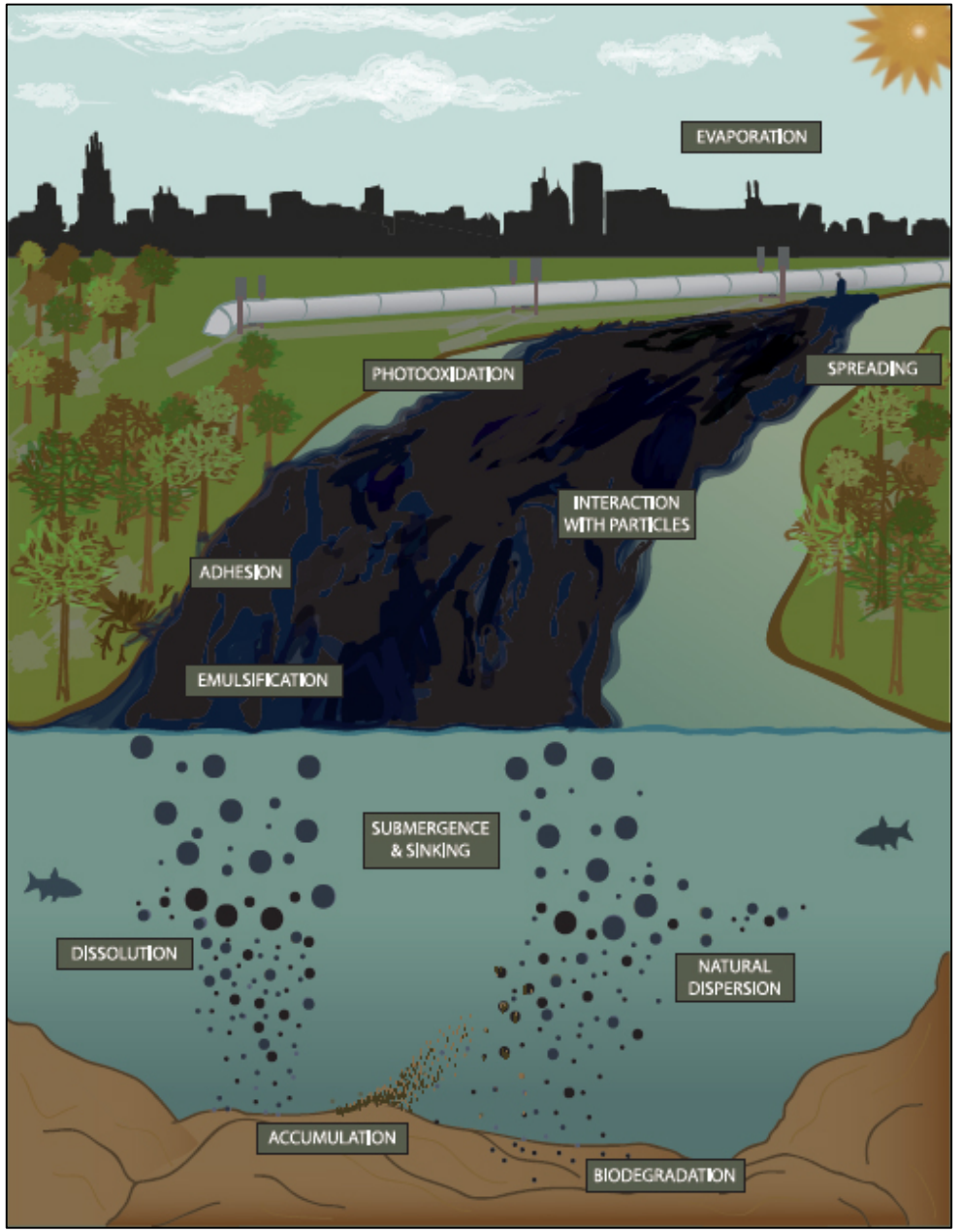



Pacific Salmon Species

# Knowledge gap about dilbit toxicity

- Dilbit, a non-conventional crude oil
  - ↓ BTEX
  - ↓ Total polycyclic aromatic hydrocarbons (PAHs)
  - ↑ 3~5 ringed PAHs in proportions
    - Most toxic and bioavailable to aquatic life

- Different environmental behaviour when spilled
  - High density and viscosity
  - High potential of sinking




Processes affecting the composition, amount, and behaviour of diluted bitumen

# Knowledge gap about dilbit toxicity

- Dilbit toxicity to fish species remains unclear
  - Very few studies on dilbit toxicity
  - Not enough empirical data

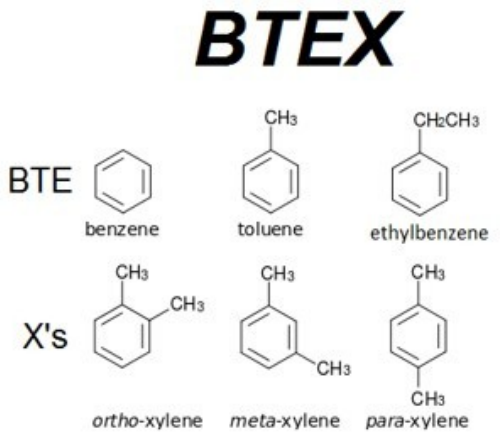
- Studies using other crude oil blend or its components (e.g. BTEX, PAHs)
  - Generalization of risk?

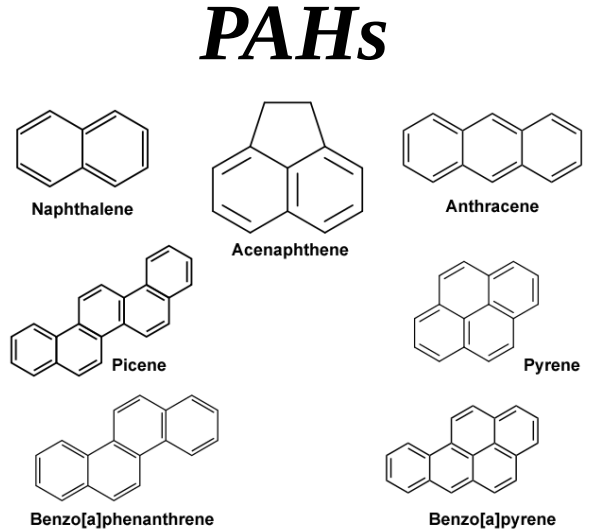
Sockeye salmon

Japanese medaka

Fathead minnow

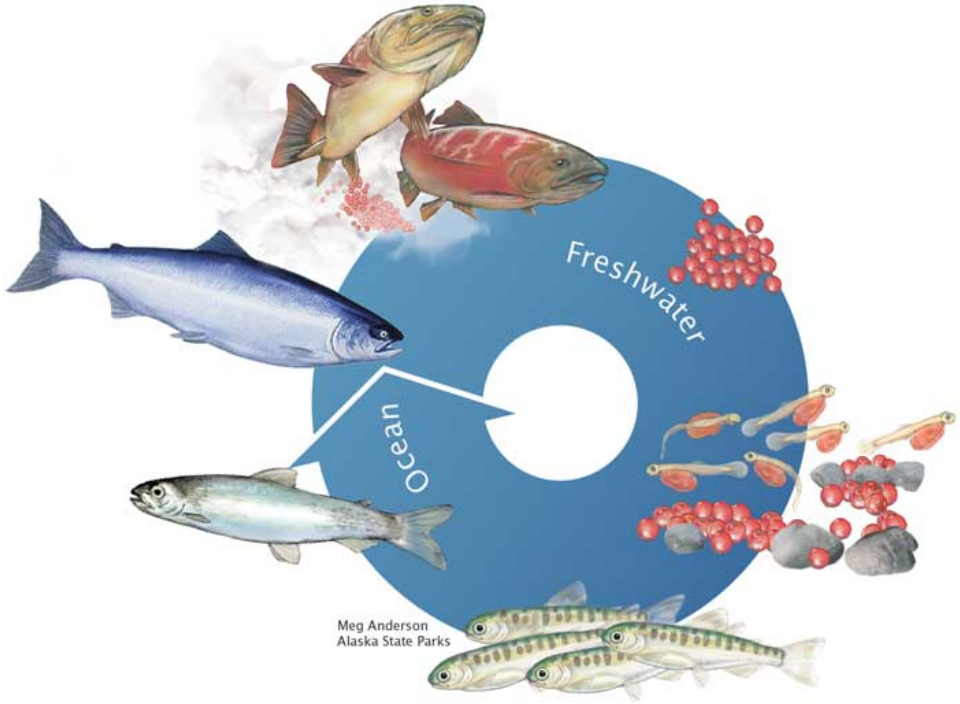
Inland silverside

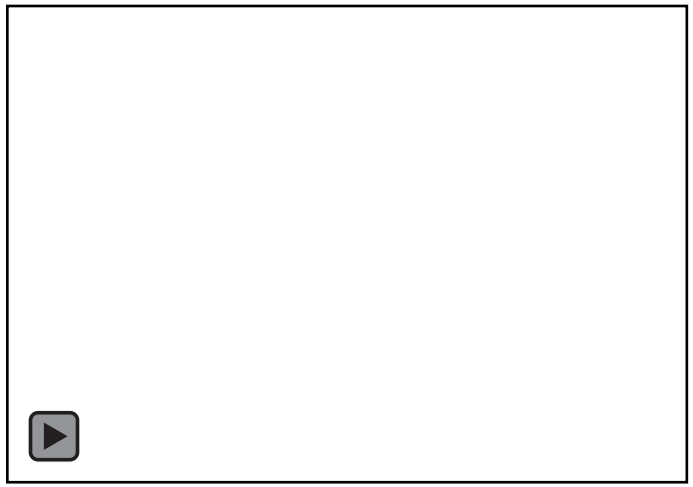


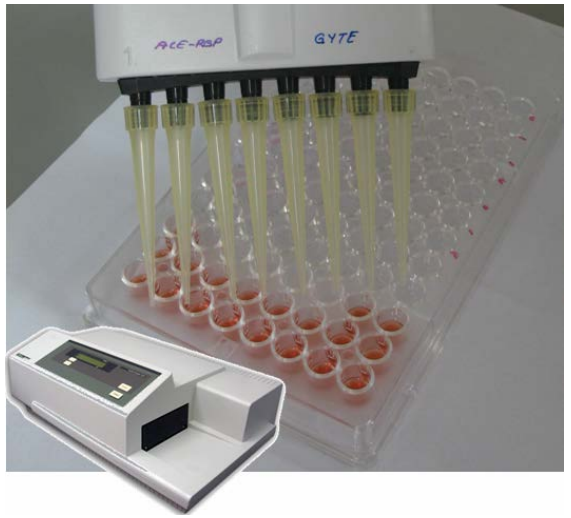



# Study objectives

- Acute and sublethal toxicity of dilbit on early life stage (ELS) Pacific salmonids
  - Mortality
  - % Deformity
  - Swimming ability
  - Biochemistry
- Latent effect in formally exposed survivors?
- Environmentally relevant exposure method



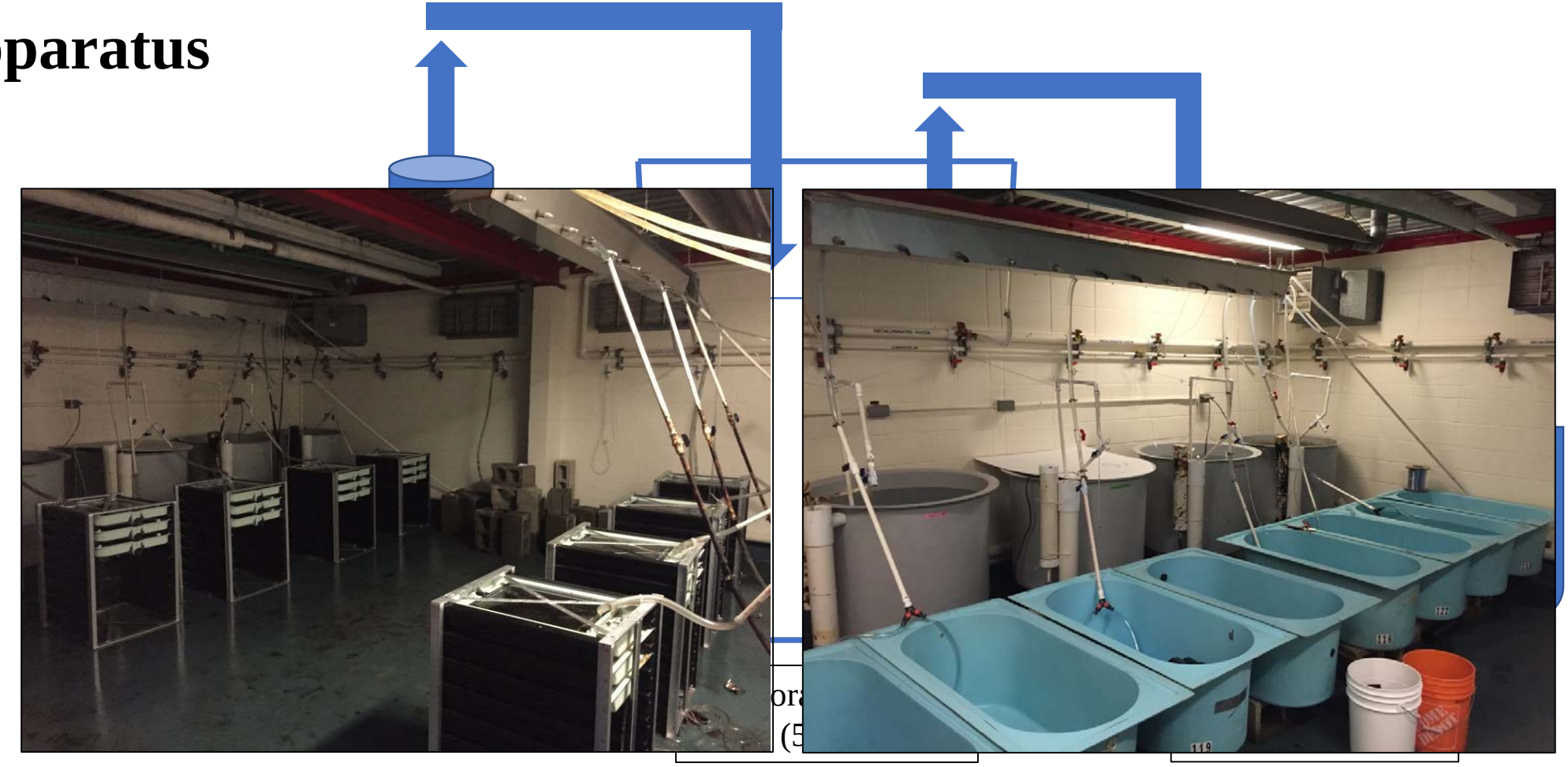


# Exposure apparatus

Siporax® ceramic beads

Dilbit

Pre-soaked for 48h



- Generate water soluble fractions (WSFs) of dilbit
- Initial spike of total PAHs at 3.5, 16.4, 66.7 µg/L
- Beads re-soaked every 21 days

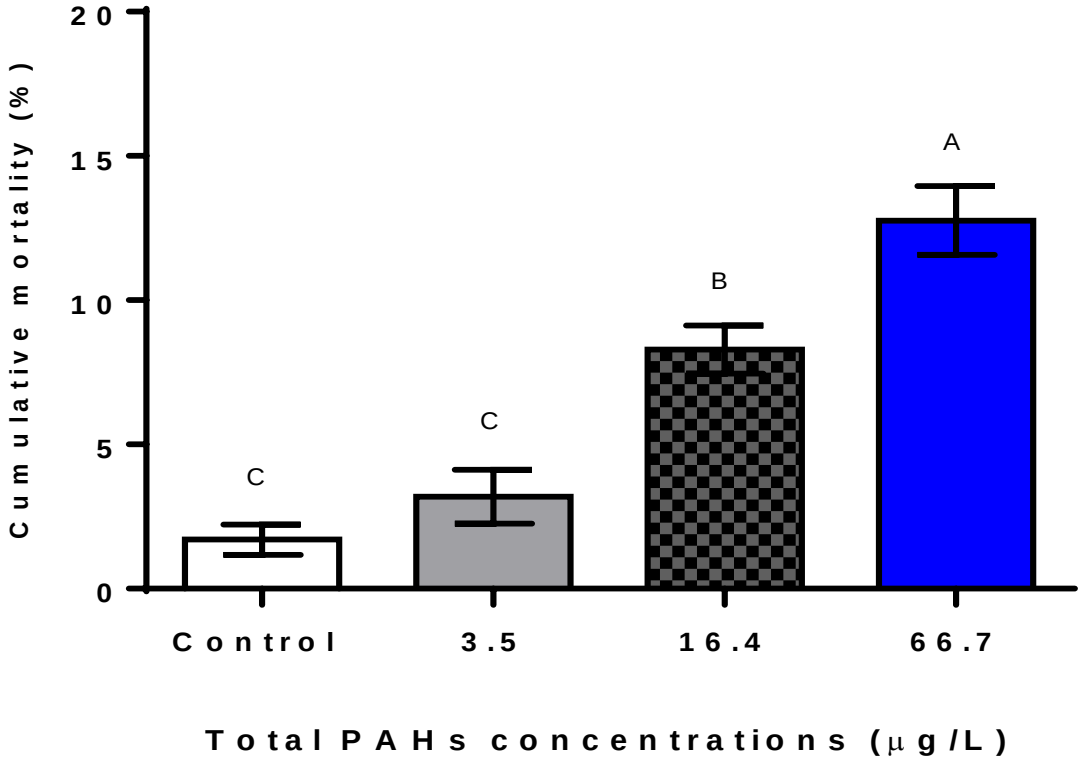
# Experiment part 1

- Fertilized sockeye salmon embryos exposed to 3 concentrations of WSFs
  - 3.5, 16.4, 66.7 µg/L total PAHs
  - Control at 0.002 µg/L
- At 50% yolk sac absorption
  - Deformity assessment
- At swim-up stage
  - Burst swimming ability
  - Body biochemistry

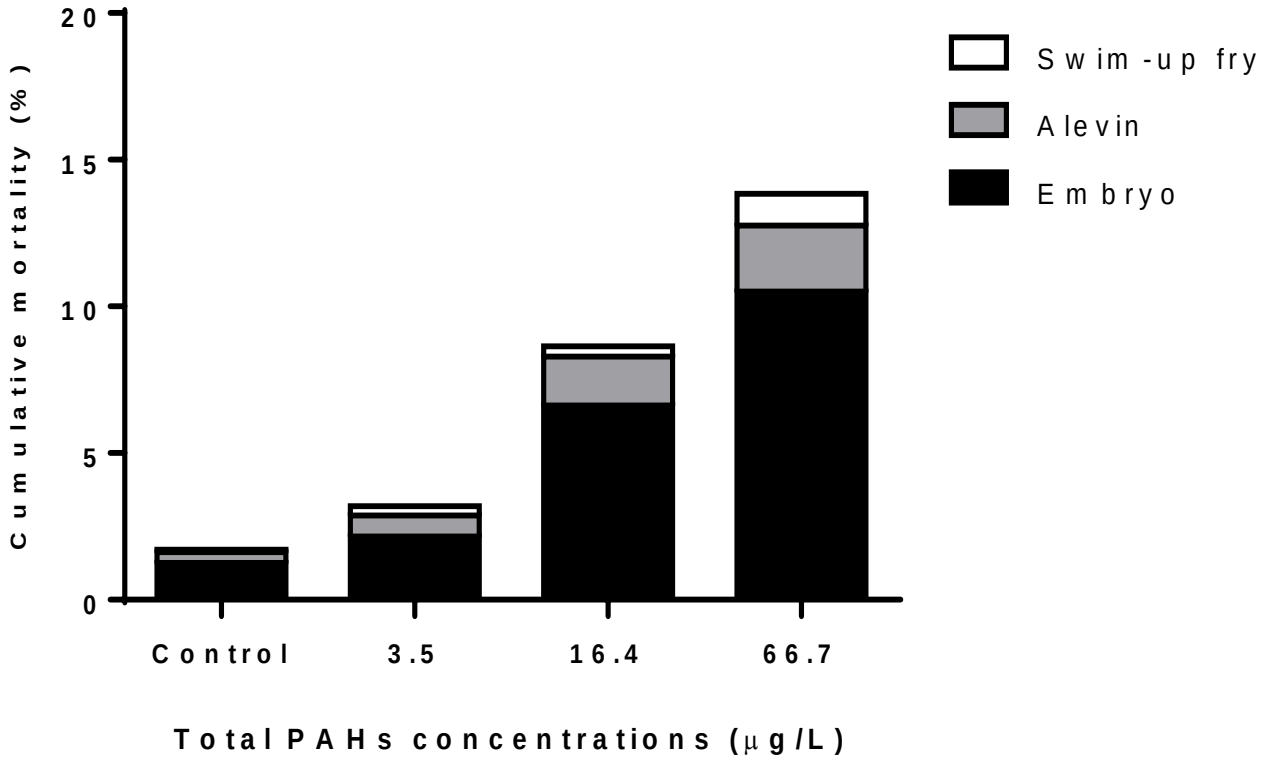
Embryo and alevin stage

Swim-up stage fry
(no external yolk sac)

# Results




Values are means ± s.e.m (n = 960). One-factor ANOVA and Tukey HSD test ($p<0.05$).



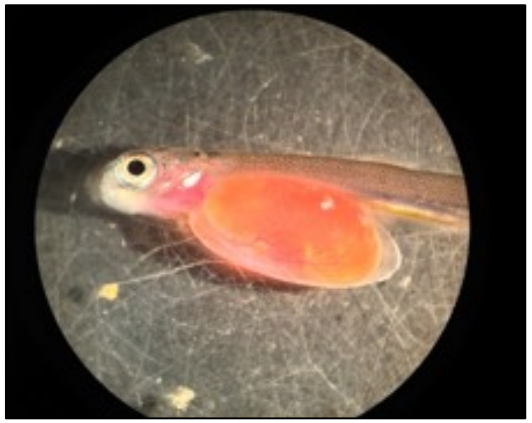

The stacked cumulative mortality (%) of fish across treatments (n = 960 ) at three different developmental stages

| | Control | 3.5 µg/L | 16.4 µg/L | 66.7 µg/L |
|---|---|---|---|---|
| Deformity (%) | | | | |
| Yolk sac edema | 2.0±1.0 **c** | 6.0±1.0 **b** | 15.0±4.0 **a** | 12.0±2.0 **a** |
| Pericardiac edema | 1.0±0 | 0 | 4.0±2.0 | 1.0±0 |
| Craniofacial | 1.0±0 | 3.0±2.0 | 3.0±1.0 | 5.0±2.0 |
| Skeletal | 0 | 1.0±0 | 1.0±0 | 1.0±0 |
| Finfold | 0 | 1.0±0 | 0 | 0 |
| Fish with at least 1 type of deformity | 2.0±1.0 **c** | 8.0±1.0 **b** | 20.0±2.0 **a** | 17.5 ±0.5 **a** |

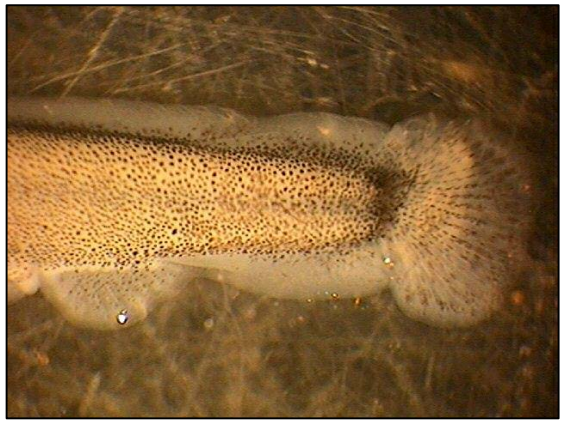
Values are means ± s.e.m (n = 200). One-factor ANOVA and Tukey HSD test ($p<0.05$)

Skeletal deformity

Craniofacial deformity



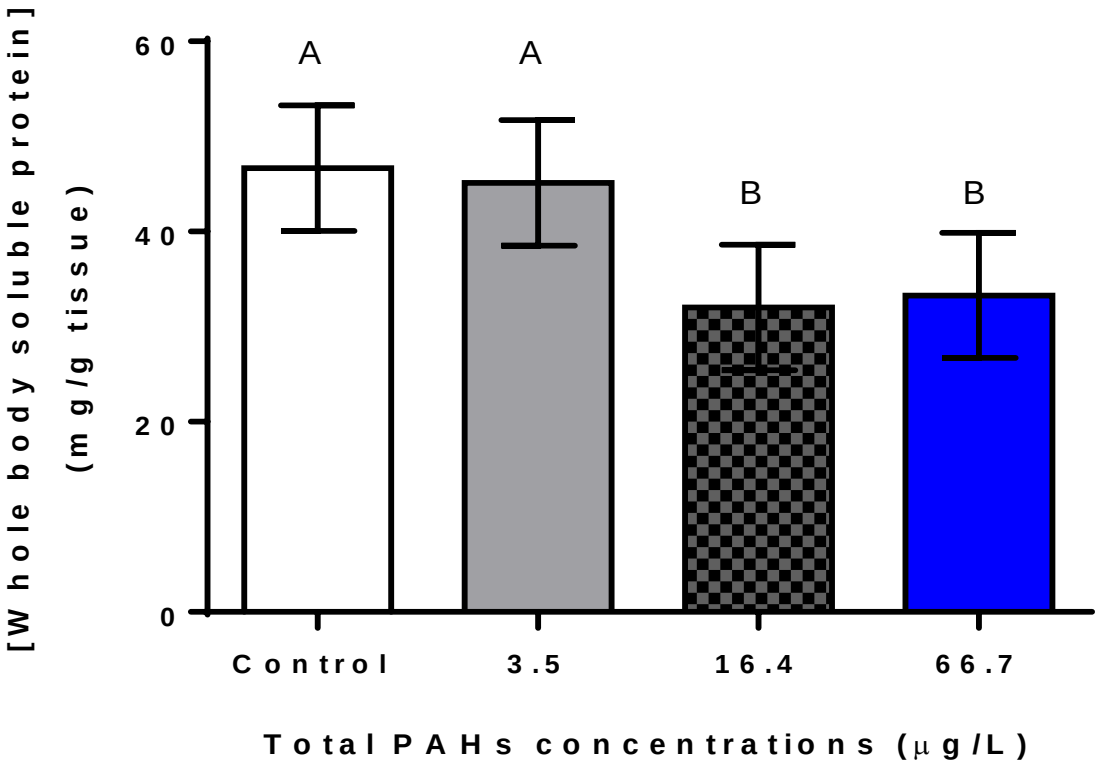
Presence of edema

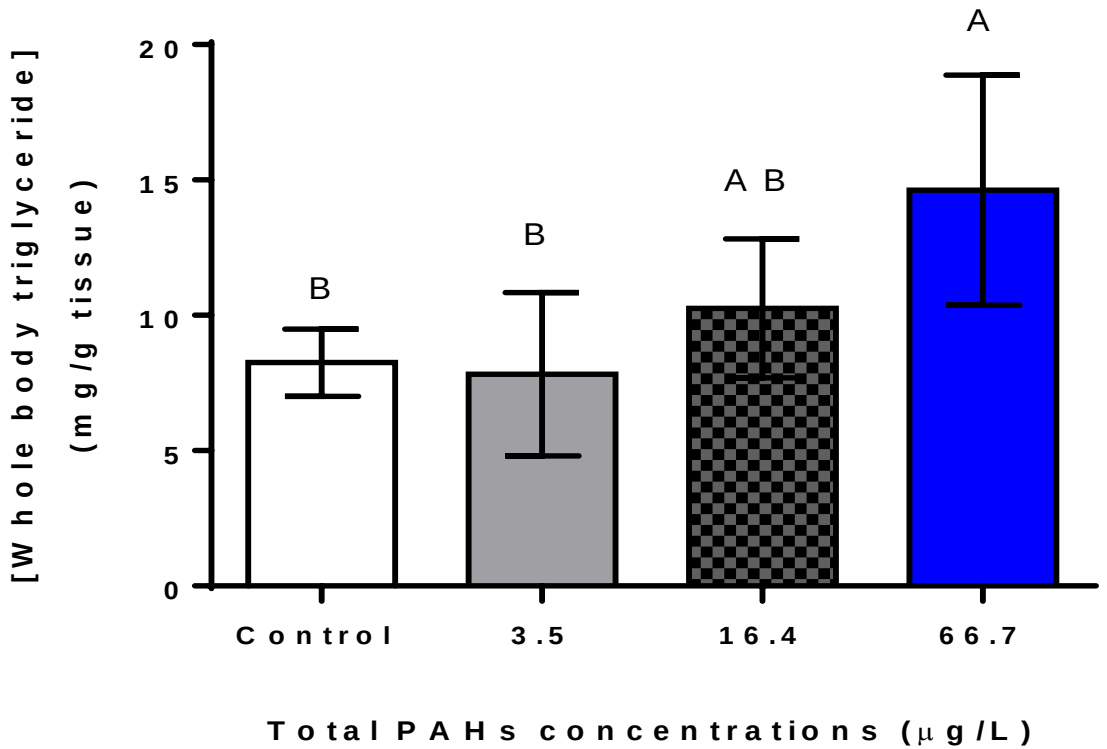


Finfold deformity

# Results

Swim-up stage fry

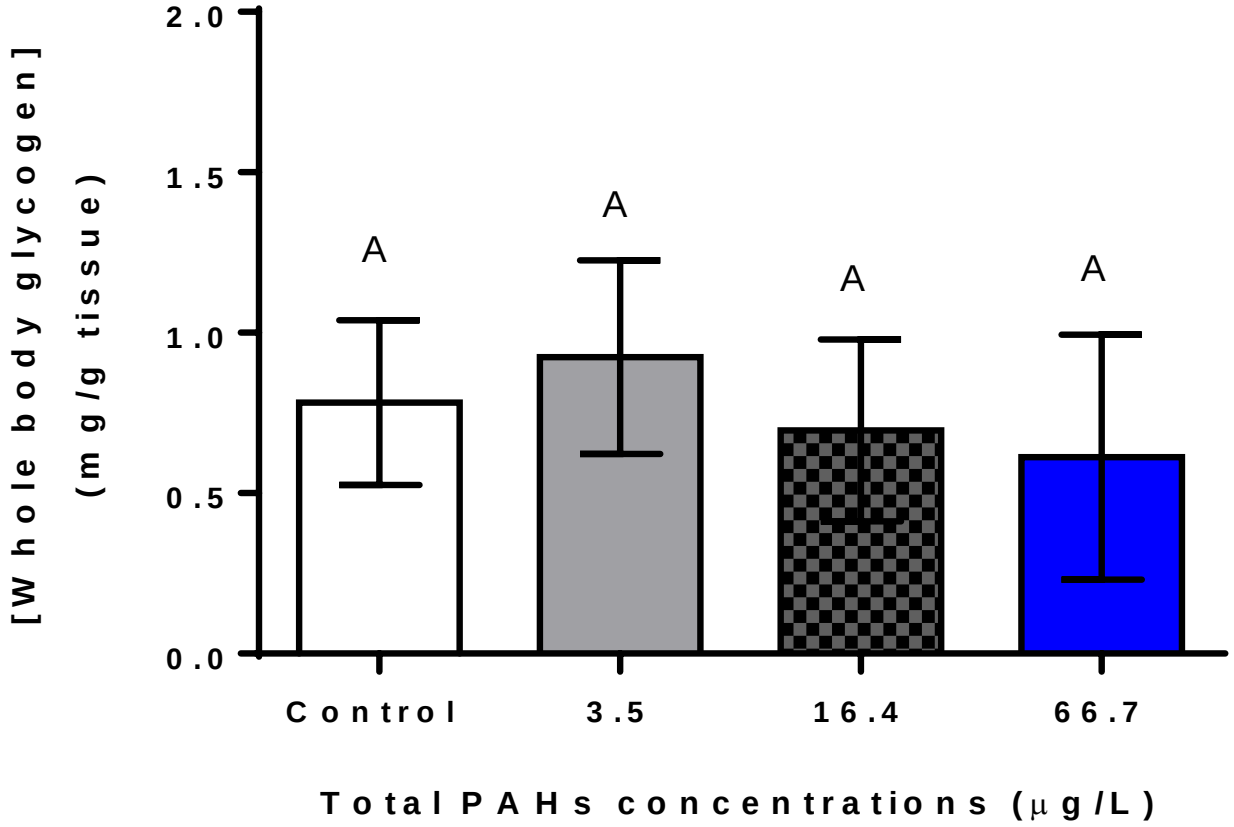



Values are means ± 95% confidence interval (n = 20). One-factor ANOVA and Tukey HSD test ($p<0.05$).

# Results

Swim-up stage fry

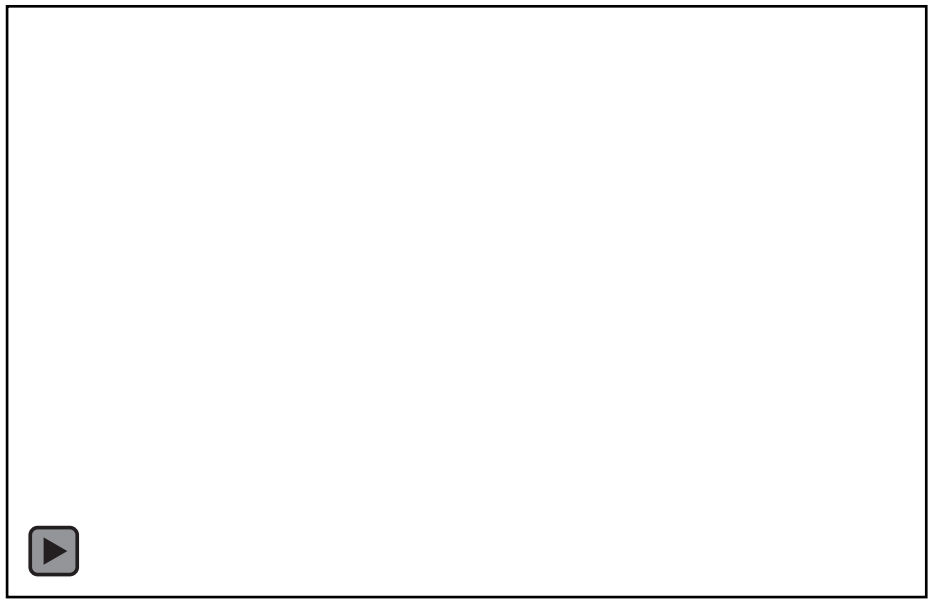



Values are means ± 95% confidence interval (n = 20). One-factor ANOVA and Tukey HSD test ($p<0.05$).

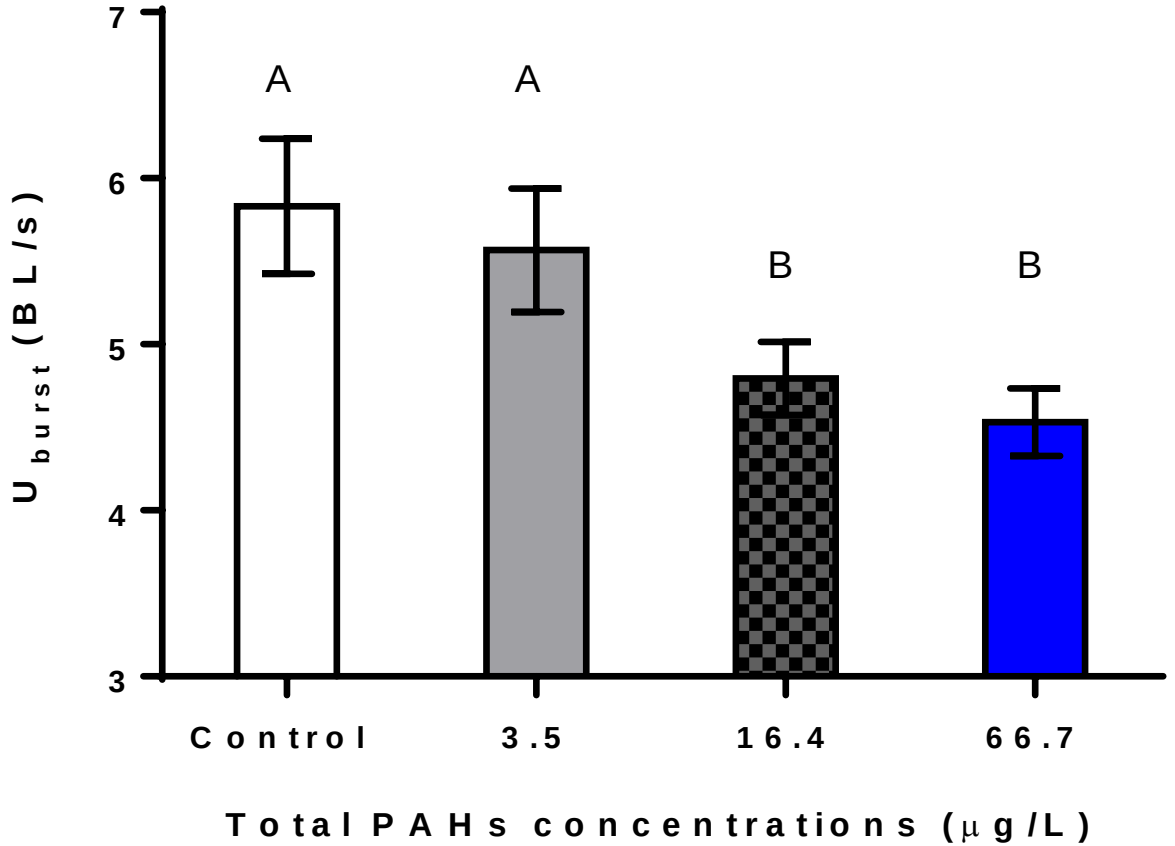
- Unchanged carbohydrate storage
- Increased lipid storage
- Decreased soluble protein

Swim-up fry

- $U_{burst}$ swimming speed --- measure of burst ability
  - Involved in food capture and predation avoidance



Loligo swimming tunnel used for $U_{burst}$ test



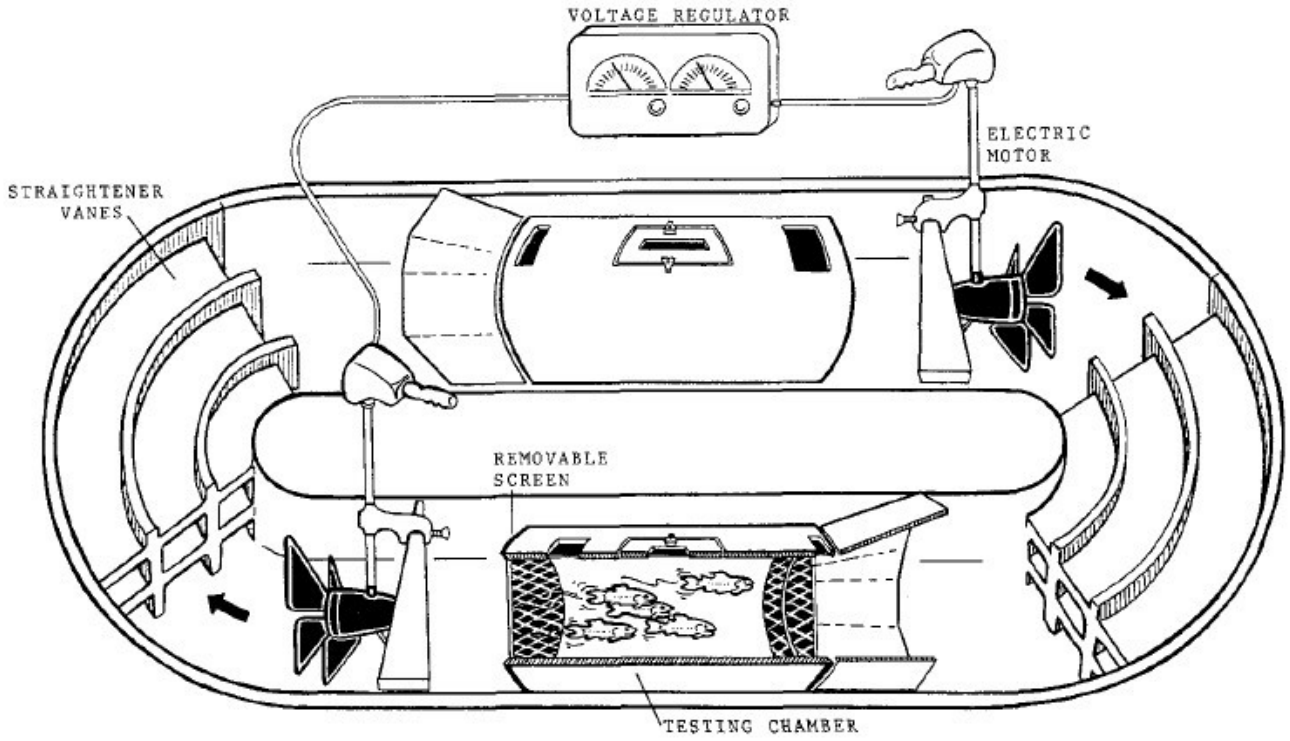

Values are means ± 95% confidence interval (n = 12). One-factor ANOVA and Tukey HSD test ($p<0.05$).

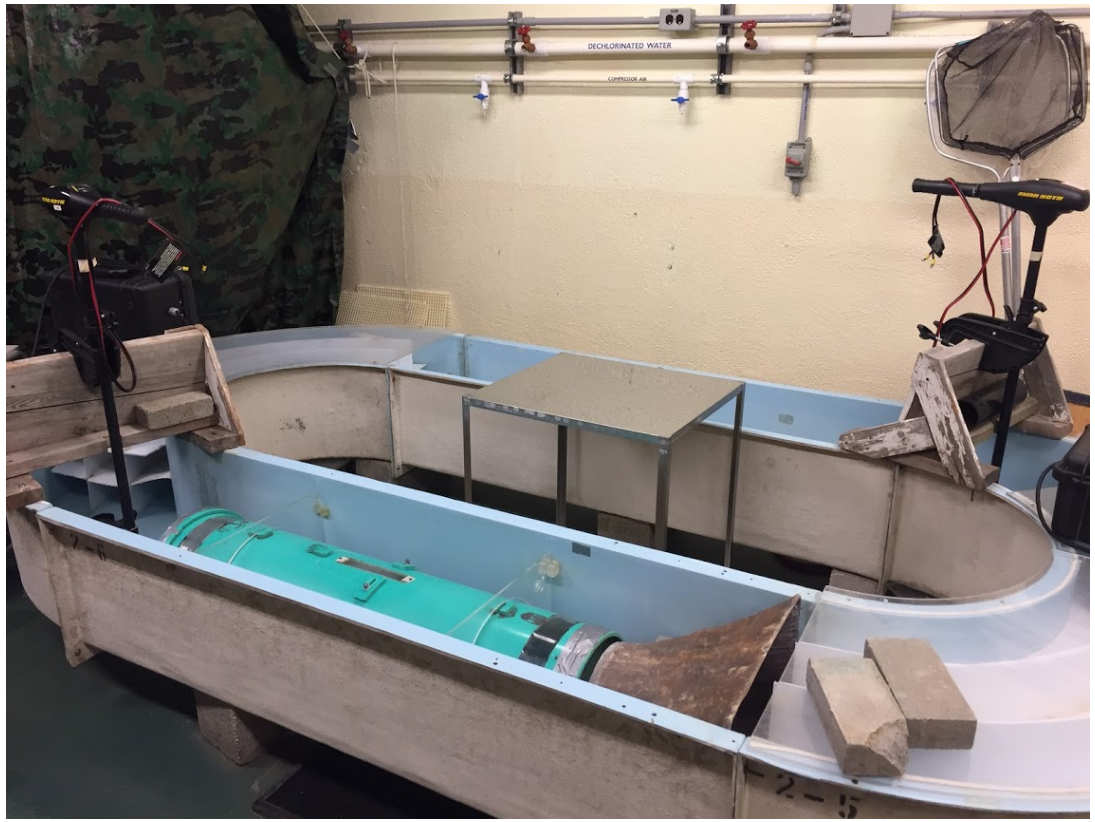
Exposed swim-up fry

Yearling parr

Pre-smolt stage

Reared in clean water for 8 months. $U_{burst}$ swiming speed tested at 1, 3, 6, or 8 months in clean water

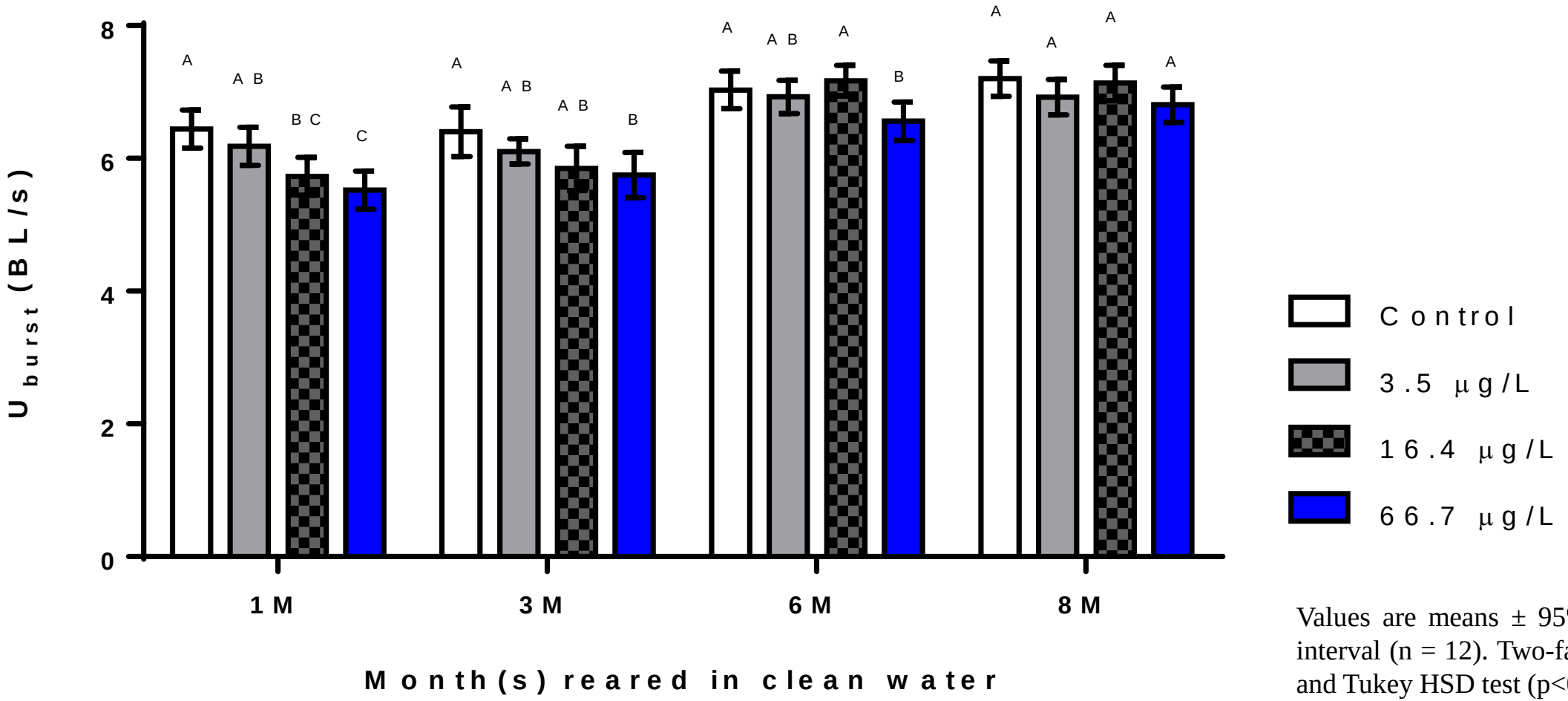


Exposed swim-up fry

Yearling parr

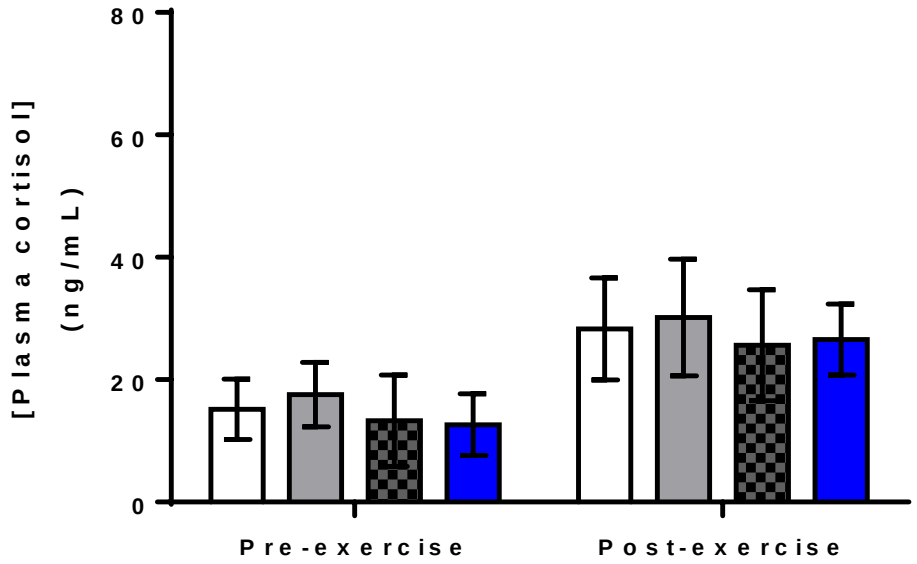
Pre-smolt stage

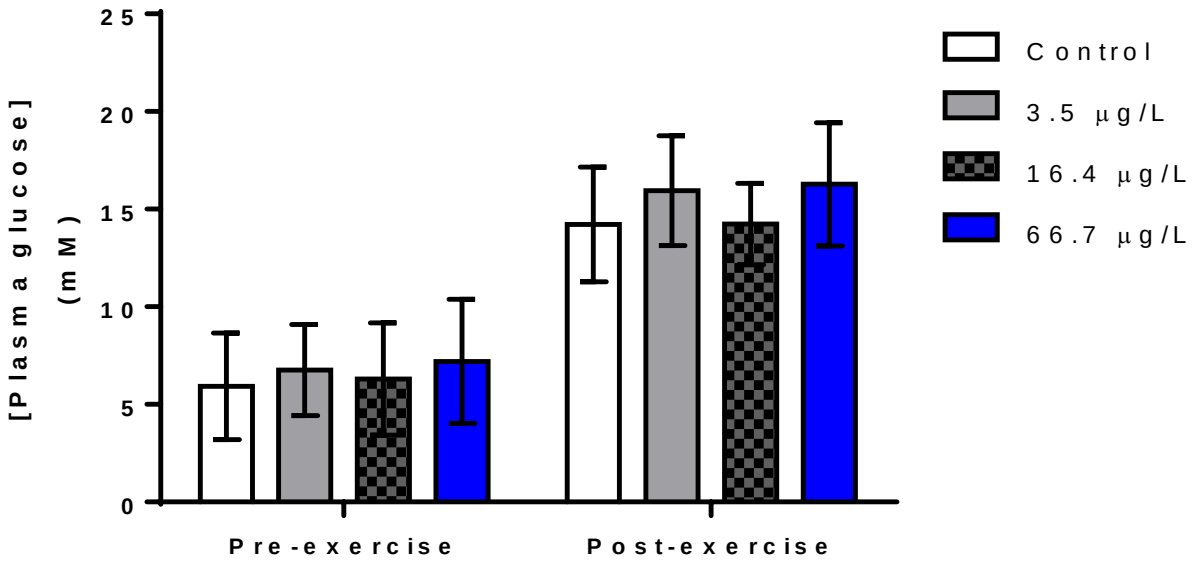


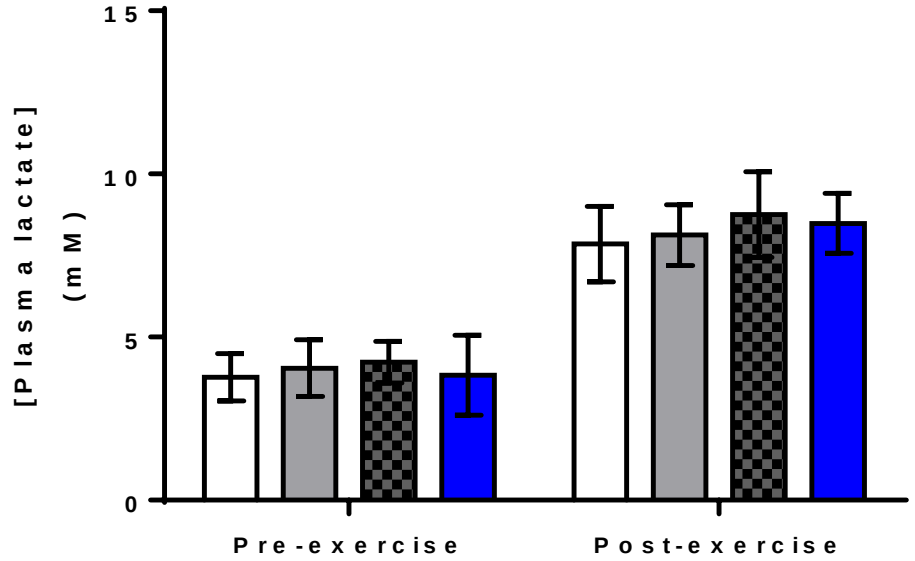

Values are means ± 95% confidence interval (n = 12). Two-factor ANOVA and Tukey HSD test ($p<0.05$).

# Pre- and post- exercise biochemistry

Values are means ± 95% confidence interval (n = 12). One-factor ANOVA and Tukey HSD test ($p<0.05$).

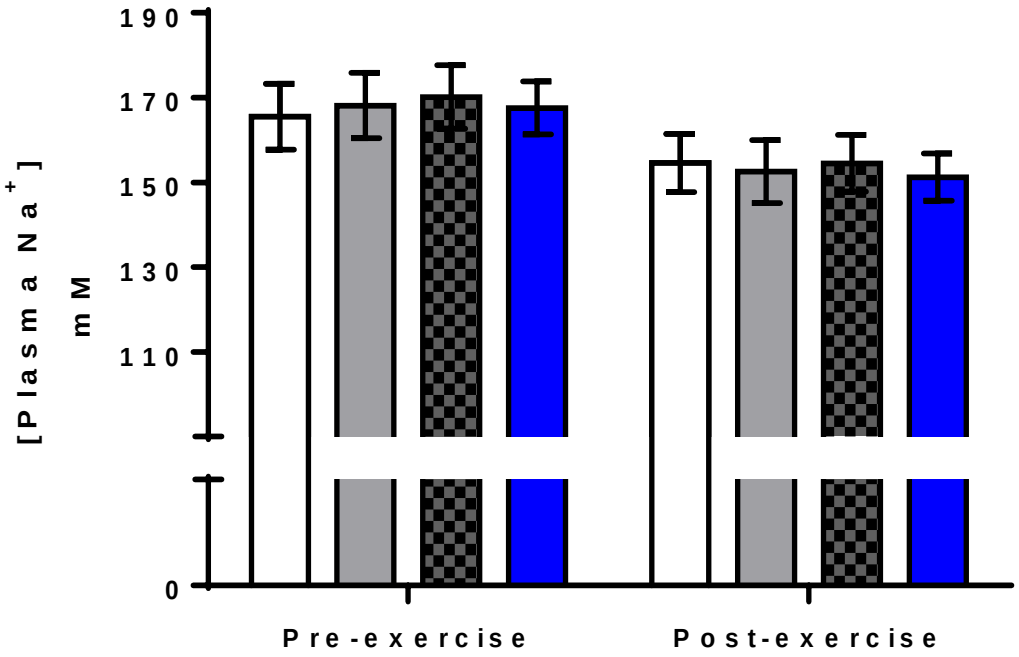


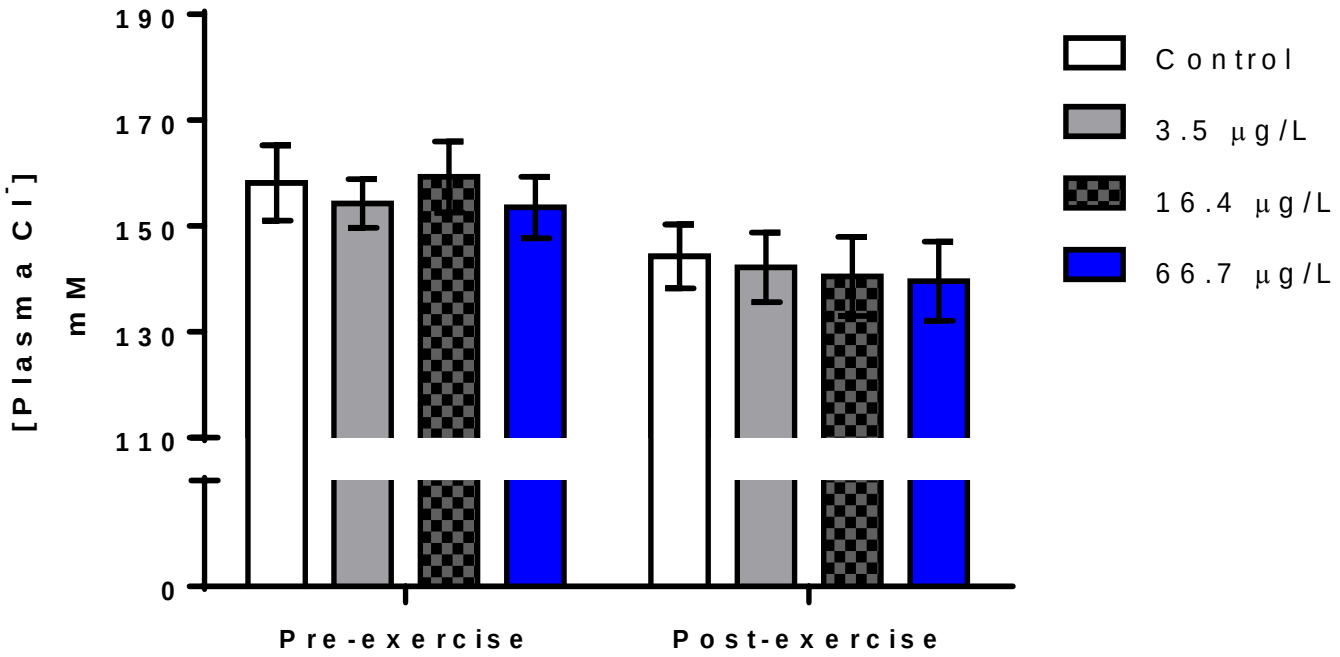




- Indicators of stress level
- Spikes in circulating cortisol, [glucose], [lactate] after exhaustive exercise

# Pre- and post- exercise biochemistry



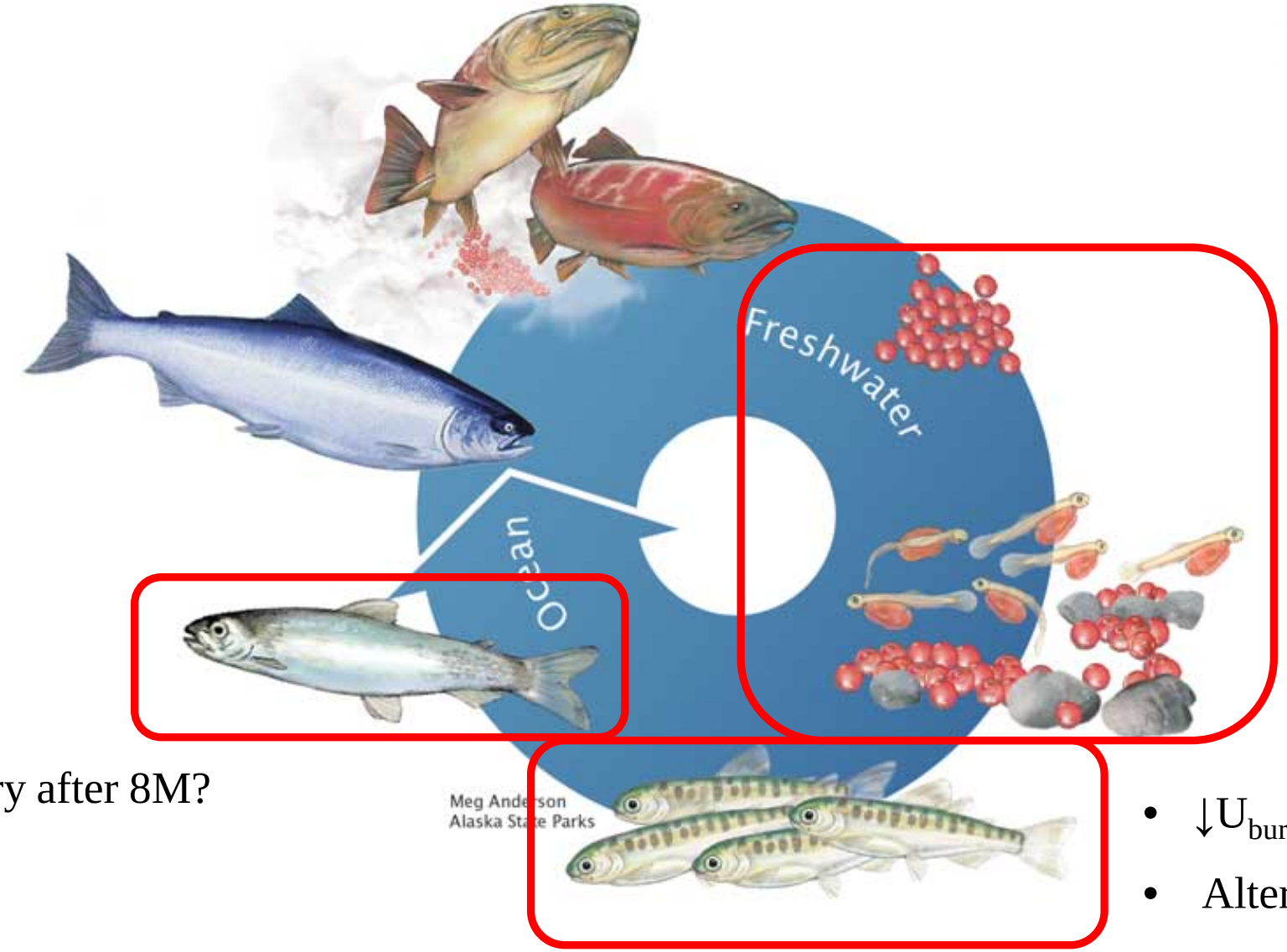




- Indicators of water-ion homeostasis
- Reduction in circulating plasma [$Na^+$] and [$Cl^-$] after exhaustive exercise

Values are means ± 95% confidence interval (n = 12). One-factor ANOVA and Tukey HSD test ($p<0.05$).

# Conclusions

## Exposure to WSFs of dilbit:




- ↑ Mortality
- ↑ % Deformity

- Latent effect
- Signs of recovery after 8M?

- ↓$U_{burst}$ swimming speed
- Altered biochemistry

# Experiment part 2 (ongoing)

Pre-smolt stage

- Exposed juvenile sockeye salmon exposed to 3 concentrations of WSFs
  - 3 exposure length: 24h, 96h, or 14d

- $U_{burst}$ and body biochemistry
  - Exercise recovery ability
  - Stress response

Exposure set-up

# Thank you for listening!

## Acknowledgements:


Fisheries and Oceans Canada

Pêches et Océans Canada

Funding support by the National Contaminant Advisory Group

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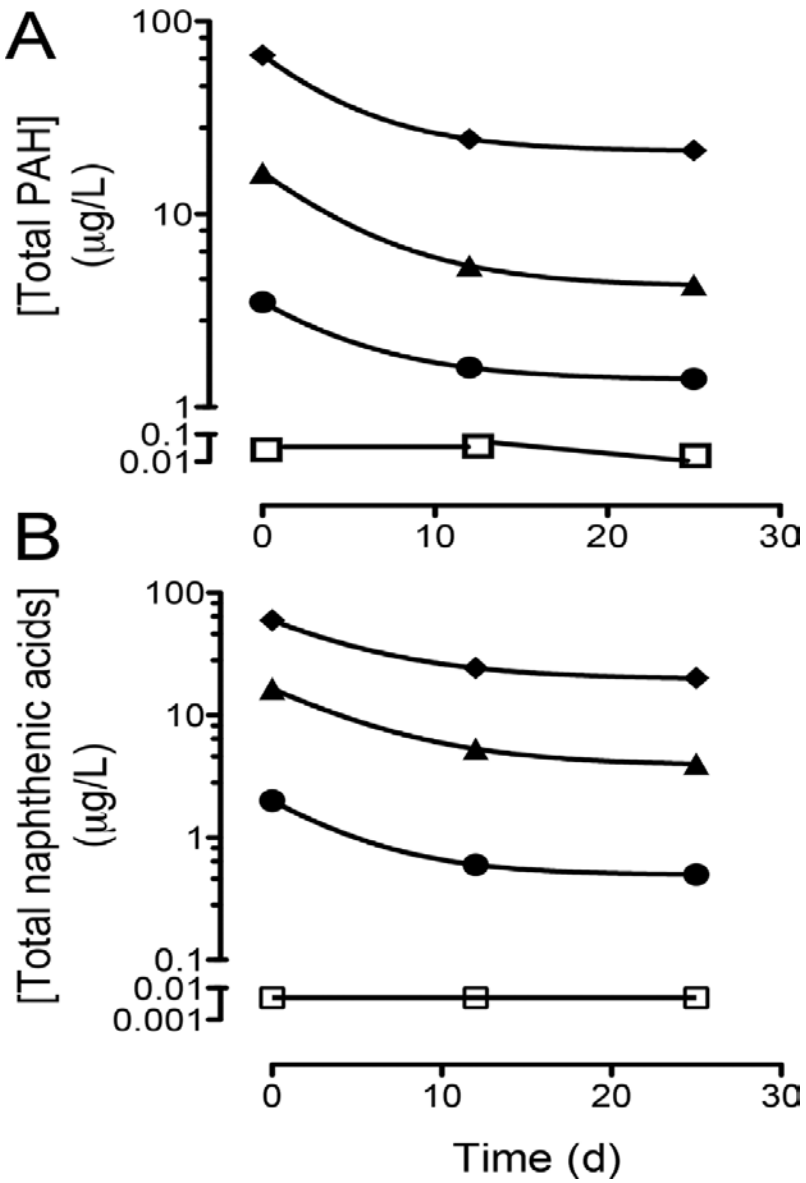
Supervision by Dr. Chris Kennedy

Animal care staff at SFU for extensive fish care


A
100
10
1
0.1
0.01
[Total PAH]
(μg/L)
0
10
20
30
B
100
10
1
0.1
0.01
0.001
[Total naphthenic acids]
(μg/L)
0
10
20
30
Time (d)

Water accommodated fractions (WAFs)